

IN THE CLAIMS:

1. (Original) A vector capable of expressing an α -1,2-mannosidase or a functional part thereof in a methylotrophic yeast strain, comprising a nucleotide sequence coding for said α -1,2-mannosidase or said functional part.

2-34. (Canceled)

35. (New) A vector for expressing an α -1,2-mannosidase or a functional part thereof in a methylotrophic yeast strain, comprising a nucleotide sequence coding for said α -1,2-mannosidase or said functional part.

36. (New) The vector of claim 35, wherein said α -1,2-mannosidase is a protein of a fungal origin or of a mammalian origin.

37. (New) The vector of claim 36, wherein said α -1,2-mannosidase is derived from *Aspergillus*, *Trichoderma reesei*, *S. cerevisiae*, murine, rabbit, or human.

38. (New) The vector of claim 35, wherein said α -1,2-mannosidase or said functional part is genetically engineered to contain an ER-retention signal.

39. (New) The vector of claim 38, wherein said ER-retention signal comprises peptide HDEL (SEQ ID NO:1).

40. (New) The vector of claim 35, wherein the nucleotide sequence coding for said α -1,2-mannosidase or said functional part is operably linked to a promoter and a 3' termination sequence, wherein said promoter is the promoter of a gene selected from the group consisting of AOXI, AOXII, GAP, and FLD.

41. (New) A vector for expressing a glucosidase II or a functional part thereof in

a methylotrophic yeast strain, comprising a nucleotide sequence coding for said glucosidase II or said functional part.

42. (New) The vector of claim 41, wherein said glucosidase II is a protein of a fungal origin or of a mammalian origin.

43. (New) The vector of claim 42, wherein said glucosidase II is *Saccharomyces cerevisiae* glucosidase II.

44. (New) The vector of claim 41, wherein said glucosidase II or said functional part is genetically engineered to contain an ER-retention signal.

45. (New) The vector of claim 44, wherein said ER-retention signal comprises peptide HDEL (SEQ ID NO:1).

46. (New) The vector of claim 41, wherein the nucleotide sequence coding for said glucosidase II or said functional part is operably linked to a promoter and a 3' termination sequence, wherein said promoter is the promoter of a gene selected from the group consisting of AOXI, AOXII, GAP, and FLD.

47. (New) A vector for disrupting the OCH1 gene in a methylotrophic yeast strain, comprising a portion of the OCH1 gene operably linked to a selectable marker gene, wherein said portion of the OCH1 gene operably linked to said selectable marker gene is capable of disrupting the OCH1 gene in said methylotrophic yeast strain.

48. (New) A genetically engineered strain of a methylotrophic yeast, wherein said strain is transformed with at least one of the vectors of claims 35, 41 or 47.

49. (New) The genetically engineered strain of claim 48, wherein said methylotrophic yeast is *Pichia pastoris*.

50. (New) A genetically engineered strain of a methylotrophic yeast, wherein said strain is transformed with a nucleotide sequence coding for an α -1,2-mannosidase or a functional part thereof, and wherein the OCH1 gene in said strain is disrupted.

51. (New) The genetically engineered strain of claim 50, wherein said methylotrophic yeast is *Pichia pastoris*.

52. (New) A genetically engineered strain of a methylotrophic yeast, wherein said strain is transformed with a nucleotide sequence coding for a glucosidase II or a functional part thereof, and wherein the OCH1 gene in said strain is disrupted.

53. (New) The genetically engineered strain of claim 52, wherein said methylotrophic yeast is *Pichia pastoris*.

54. (New) A method of reducing glycosylation on proteins produced from a methylotrophic yeast, comprising transforming said yeast with at least one of the vectors of claims 35, 41 or 47.

55. (New) The method of claim 54, wherein said yeast is *Pichia pastoris*.

56. (New) A method of reducing glycosylation on proteins produced from a methylotrophic yeast, comprising transforming said yeast with a nucleotide sequence coding for an α -1,2-mannosidase or a functional part thereof, and with a nucleotide sequence comprising a portion of the OCH1 gene of said yeast operably linked to a selectable marker to effect the disruption of the OCH1 gene in said methylotrophic yeast.

57. (New) The method of claim 56, wherein said yeast is *Pichia pastoris*.

58. (New) A method of reducing glycosylation on proteins produced from a

methylophilic yeast, comprising transforming said yeast with a nucleotide sequence coding for a glucosidase II or a functional part thereof, and with a nucleotide sequence comprising a portion of the OCH1 gene of said yeast operably linked to a selectable marker to effect the disruption of the OCH1 gene in said methylophilic yeast.

59. (New) The method of claim 58, wherein said yeast is *Pichia pastoris*.

60. (New) A method of reducing glycosylation of a heterologous glycoprotein expressed from a methylophilic yeast, comprising transforming cells of said methylophilic yeast with at least one of the vectors of claims 35, 41 or 47, and producing said glycoprotein from the transformed cells.

61. (New) The method of claim 60, wherein said yeast is *Pichia pastoris*.

62. (New) A method of reducing glycosylation of a heterologous glycoprotein expressed from a methylophilic yeast, comprising transforming cells of said yeast with a nucleotide sequence coding for an α -1,2-mannosidase or said functional part thereof, and with a nucleotide sequence comprising a portion of the OCH1 gene of said yeast operably linked to a selectable marker, such that said α -1,2-mannosidase or said functional part thereof is expressed in transformed cells, and the OCH1 gene in said methylophilic yeast is disrupted; and producing said glycoprotein from the transformed cells.

63. (New) The method of claim 62, wherein said yeast is *Pichia pastoris*.

64. (New) A method of reducing glycosylation of a heterologous glycoprotein expressed from a methylophilic yeast, comprising transforming cells of said yeast with a nucleotide sequence coding for a glucosidase II or a functional part thereof, and with a nucleotide sequence comprising a portion of the OCH1 gene of said yeast operably linked to a selectable marker, such that said glucosidase II or said functional part thereof is expressed in transformed cells, and the OCH1 gene in said methylophilic yeast is disrupted; and producing said

glycoprotein from the transformed cells.

65. (New) The method of claim 64, wherein said yeast is *Pichia pastoris*.
66. (New) A glycoprotein produced by the method of claim 60.
67. (New) A glycoprotein produced by the method of claim 62.
68. (New) A glycoprotein produced by the method of claim 64.
69. (New) A kit comprising at least one of the vectors of claims 35, 41 or 47.
70. (New) The kit of claim 69, further comprising a methylotrophic yeast strain.
71. (New) A kit comprising the methylotrophic yeast strain of claim 48.
72. (New) A kit comprising the methylotrophic yeast strain of claim 50.
73. (New) A kit comprising the methylotrophic yeast strain of claim 52.
74. (New) A method for producing in methylotrophic yeast, glycoproteins having carbohydrate structures similar to those produced by human cells, comprising providing a methylotrophic yeast strain, which does not express at least one enzyme involved in production of high mannose structures; and introducing into the yeast strain at least one enzyme for production of Man₅GlcNAc₂.
75. (New) The method of claim 74, wherein said enzyme involved in production of high mannose structures is alpha-1,6-mannosyltransferase encoded by the OCH1 gene.
76. (New) The method of claim 74, wherein said methylotrophic yeast strain is

an OCH1 mutant strain.

77. (New) The method of claim 76, wherein said OCH1 mutant strain is made by transforming a wild type methylotrophic yeast strain with the vector of claim 47.

78. (New) The method of claim 74, wherein said enzyme for production of $\text{Man}_5\text{GlcNAc}_2$ is a mannosidase or glucosidase.

79. (New) The method of claim 78, wherein said mannosidase is α -1,2-mannosidase.

80. (New) The method of claim 78, wherein said glucosidase is glucosidase II.

81. (New) The method of claim 74, wherein said enzyme for production of $\text{Man}_5\text{GlcNAc}_2$ is of a fungal origin or a mammalian origin.

82. (New) The method of claim 74, wherein said enzyme for production of $\text{Man}_5\text{GlcNAc}_2$ is targeted to a subcellular location in said methylotrophic yeast where it is optimal to produce $\text{Man}_5\text{GlcNAc}_2$.

83. (New) The method of claim 82, wherein said subcellular location is the ER.

84. (New) The method of claim 74, wherein said methylotrophic yeast is of the genera *Candida*, *Hansenula*, *Torulopsis*, or *Pichia*.

85. (New) The method of claim 84, wherein said methylotrophic yeast is selected from *Pichia pastoris*, *Pichia methanolica*, *Pichia anomola*, *Hansenula polymorpha* or *Candida boidinii*.

86. (New) A method for producing in methylotrophic yeast, glycoproteins having carbohydrate structures similar to those produced by human cells, comprising providing a methylotrophic yeast strain, which does not express at least one enzyme involved in production of high mannose structures; and introducing into the yeast strain at least one enzyme for production of $\text{Man}_5\text{GlcNAc}_2$, wherein said enzyme for production of $\text{Man}_5\text{GlcNAc}_2$ is targeted to a subcellular location in said methylotrophic yeast where it is optimal to produce $\text{Man}_5\text{GlcNAc}_2$.

87. (New) A method for producing in methylotrophic yeast, glycoproteins having carbohydrate structures similar to those produced by human cells, comprising providing a methylotrophic yeast strain, which does not express at least one enzyme involved in production of high mannose structures; and introducing into the yeast strain at least one enzyme for production of $\text{Man}_5\text{GlcNAc}_2$, wherein said enzyme for production of $\text{Man}_5\text{GlcNAc}_2$ is targeted to a subcellular location in said methylotrophic yeast and wherein said subcellular location is the ER.

88. (New) A method for producing in a methylotrophic yeast, glycoproteins having carbohydrate structures similar to those produced by human cells, comprising introducing into the yeast at least one enzyme for the production of $\text{Man}_5\text{GlcNAc}_2$, and producing said glycoproteins in said yeast.

89. (New) A method for producing in methylotrophic yeast, glycoproteins having carbohydrate structures similar to those produced by human cells, comprising providing a methylotrophic yeast strain which does not express at least one enzyme involved in production of high mannose structures, and producing said glycoproteins in said strain.